

Figure 2: Sequence comparison of the $\Delta 4$ -desaturases from *Euglena gracilis* and *Thraustochytrium* (WO 200226946). The two sequences show 35% identity [CLUSTAL W(1.60) multiple sequence alignment]

<i>Euglena</i>	MLVLFGNFYVKQYSQKNGKPENGATPENGAKPPCENGTVKEKRENDTANVRPTRPAGPPP
<i>Thrausto</i>	-----
<i>Euglena</i>	ATYYDSLAVSGQGERLFTTDEVRRHILPTDGLWLTCHGVYDVTDFLAKHPGGGVITLGL
<i>Thrausto</i>	-----MTVGDEEIPFEQVRAHNKPPDDAWCAIHGHVYDVTKFASVHPGGDIILLAA
<i>Euglena</i>	GRDCTILIESYHPAGRPDKVMEKYRIGTLQDP-----KTFYAWGESDFY
<i>Thrausto</i>	GKEATVLYETYHVRGVSDAVLRKYRIGKLPDQGGANEKEKRTL SGLSSASYTTWNSDFY
<i>Euglena</i>	PELKRRALARLKEAGQARRG--GLGVKALLVLTFFVSWYMWVAHKS-----FLWAAVWGF
<i>Thrausto</i>	RVMRERVVARLKERGKARRGGYELWIKAFLLLVGFWSLSLYMCTLDPSFGAILAAMSLGV
<i>Euglena</i>	AGSHVGLSIQHDGNHGAFSRNTLVNRLAGWMDLIGASSTVWEYQHVIGHHGYTNLVS--
<i>Thrausto</i>	FAAFVGTCIQHDGNHGAFASQSRVWNVKAVAGWTLDMIGASGMTWEFQHVLGHHPYTNLIEEE
<i>Euglena</i>	-----DTLFLSLPENDPDVFSYPLMRMHPDTAWQPHHRFQHLFAFPLFALMTIS
<i>Thrausto</i>	NGLQKVSQKKMDTKLADQESDPDVFTYPMNRLHPWHQKRWHYHRFQHIYGPFIFGFMTIN
<i>Euglena</i>	KVLTSDFAVCLSMKKGSIDCSSRLVPLEGQLFWGAKLANFLQLIVLPCYLHGTAMGLAL
<i>Thrausto</i>	KVVTQDVGVVLRKRLFQIDAECRYASPMYVARFWIMKALTVLYMVVALPCYMQGPWHGLKL
<i>Euglena</i>	FSVAHLVSGEYLAICFIINHISECFMN-----TSFQTAARRTEM LQAAHQQA
<i>Thrausto</i>	FAIAHFTCGEVLATMFIVNHHIEGVSYASKDAVKGTMAPPKTMHGVTPTMNNTRKEVEAEA
<i>Euglena</i>	AEAKKVKPTPPENDWAVTQVQCVCNWRSGGVLANHL SGGNLHQIEHHLFPSISHANYPTI
<i>Thrausto</i>	SKSGAVVKSVP LDDWAVVQCQTSVNWSVGSFWNNHFSGGNLHQIEHHLFPGLSHETYHYHI
<i>Euglena</i>	APVVKEVCEEYGLPYKNVVTFFWDAVCGMVQHLRLMGAPPVPTNGDKKS-
<i>Thrausto</i>	QDV FQSTCAEYGVYPYQHEPSLWTAYWKMLEHLRLQLGNEETHESWQRAA-

Gap Weight:	8	Average Match:	2.912	Length Weight:	2	Average Mismatch:	-2.003
Quality:	926	Length:	590	Ratio:	1.781	Gaps:	9

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|= IDENTITY;
:= 2;
.= 1
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51 RPTRPAGPPATYYDSLAVSGQKERLFTTDEVRRHILPTDGLWLTCEGV 100
      | : ::||| | | | | | |
1  .....MTVGYDEEIPFEQVRAHNKPDPAWCAIHGV 31
      . . . . .
101 YDVTDFLAKHPGGGVITLGLGRDCTILIESYHPAGRPDKVMEKYRIGTLQ 150
      |||| | . |||| : | | :: ||| | | | : |||| |
32 YDVTKFASVHPGGDIILLAAGKEATVLYETYHVGVSDAVLRKYRIGKLP 81
      . . . . .
151 DPK.....TFYAWGESDFYPELKRRALARLKEAGQARR 183
      | . ::||| | : | .||| | .|||
82 DGQGGANEKEKRTLGLSSLASYYTW.NSDFYVRMRVRVARLKERGKARR 130
      . . . . .
184 GG..LGVKALLVLTFFVSWY.MWVAHKSF...LWAAVWGFAGSHVGLSI 227
      || | :|| |. | | | | | | | | | | | |

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131 GGYELWIKAFLLLVGFWSLYWMCTLDPSFGAILAAMSLGVFAAFVGTCTI 180
228 QHDCNHGAFSRNTLVNRLAGWMDLIGASSTVWEYQHVIGHHQYTNLVS. 276
|||||... ||:.||| :||:|||| ||:||||| ||||:
181 QHDCNHGAFQAQRWVNKVAGWTLDMIGASGMTWEFQHVLGHPYTNLIEE 230
277DTLFSLPENDPDVFSSYPLMRMHPDTAWQPHHRFQHLF 314
|| : |||||||.||:|||| : ||||:|
231 ENGLQKVSQKMDTKLADQESDPDVFSTYPMRLHPWHQKRWYHRFQHIY 280
315 AFPLFALMTISKVLTSDFAVCLSMKKGSIDCSSRLVPLEGQLLFWGAKLA 364
:| |||.||. | | : | | | | | | | |
281 GPFIFGFMNTINKVVTQDVGVLKRKLFQIDAECRYASPMYVARFWIMKAL 330
365 NELLQIVLPCYLHGTMGLALFSVAHLVSGEYLAICFIINHISESCEFMN 414
| : |||: | || ||:| | | | | | | | : |
331 TVLYMVALPCYMQGPWHGLKLFALIAHFTCGEVLATMFIVNHHIEGVSYS 380
415 TSF.....QTAARRTEMLQAAHQA.AEAKK...VKPTPPNDWAVTQ 452
.| | | : ||| | | . | | | |
381 KDAVKGTMAPPKTMHGVTPMNTRKEVEAEASKGAVKSVPLDDWAVVQ 430
453 VQCCVNRSGGVLANHLSSGLNHQIEHHLFPSISHANYPTIAPVVKVECE 502
| ||| | || ||||| | | | : | | | | | |
431 CQTSVNVSVGSWFVNHFSGLNHQIEHHLFPGLSHETYHYHQDVQSTCA 480

503 EYGLPYKNYVTFWDAVCGMVQHRLRLMGAPPVPTNGDKKS* 542

|||.||.. . | | |.:||| :| . : .|

481 EYGVPIQHEPSLWTAYWKMLEHLRQLGNEETHESWQRAA* 520

Figure 4: GC analyses of yeast cells fed with DPA (docosapentaenoic acid). Control: yeast cells without $\Delta 4$ -desaturase [A], conversion to DHA in cells having the $\Delta 4$ -desaturase (pYES-EGD4-2) [B]

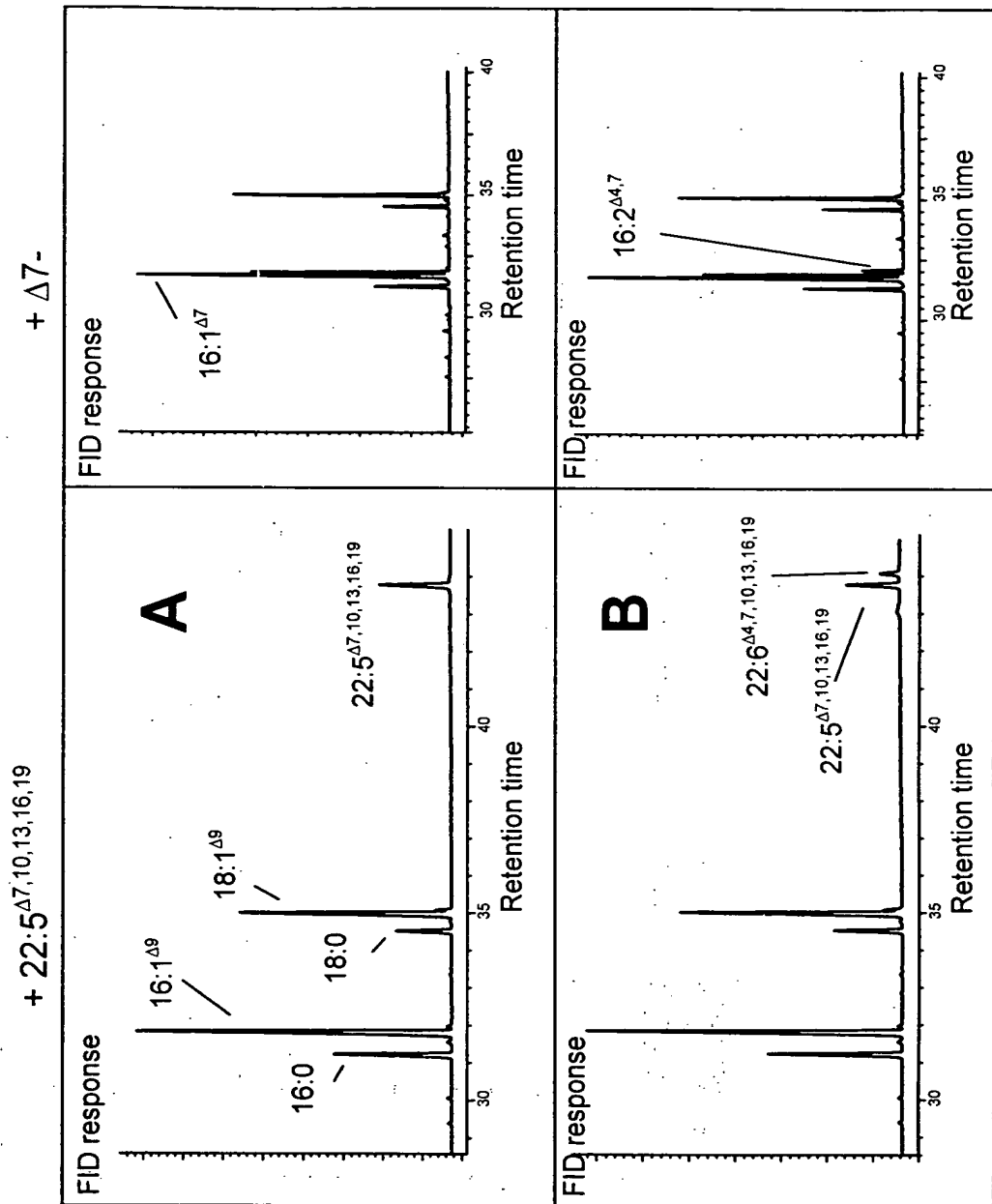


Figure 5: Position analysis of $\Delta 4$ -desaturated fatty acids. 16:1 $\Delta 7$ and 22:4 $\Delta 7, 10, 13, 16$ were used for feeding. Conversion is stated in percent.

acyl group	mol % in position	
	sn-1	sn-2
16:0	31.3	4.9
16:1 $\Delta 9$	6.5	3.5
16:2	x 4	0.6
18:0	3.6	3.3
18:1 $\Delta 9$	2.5	11.0
18:1 $\Delta 11$	0.6	1.1

acyl group	mol % in position	
	sn-1	sn-2
16:0	7	0.3
16:1 $\Delta 9$	24.2	22.2
18:0	3.5	0
18:1 $\Delta 9$	12.2	23.8
18:1 $\Delta 11$	0.8	0.7
22:4	x 10	2.2
22:5	x 10	0.1
		0.5
		2.4